CU AND ZN KINETICS IN *MYRIOPHYLLUM HETEROPHYLLUM* MICHX. AND *POTAMOGETON RICHARDSONII* (AR. BENN.) RYDB.[1]

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Abstract. Two macrophytes, *Myriophyllum heterophyllum* and *Potamogeton richardsonii*, and sediments were transplanted (1) from a site with low trace metal concentrations in the sediments to one with higher levels, and (2) from the site with high concentrations to artificial ponds containing sediments with lower concentrations to assess the uptake and retention processes for Cu and Zn.

Uptake of Cu and Zn by the sediments was extremely slow over the 100-d study; only the slope of the uptake of Zn in the upper 5 cm was significantly >0 ($P < .01$).

Nonlinear least squares fits of *P. richardsonii* and *M. heterophyllum* data to mathematical models revealed that predicted concentrations and measured uptake values were in good agreement. Uptake and retention half-times averaged 3.7 and 6.7 d, respectively, and showed little difference between plant species or trace metals. Calculated ratios lead us to speculate that known levels of Cu and Zn in one plant species may be useful in estimating concentrations in the other.

The data suggest that roots, rather than shoots, are the predominant site of uptake for Cu and Zn in these species.

Key words: copper; kinetics; Myriophyllum heterophyllum; Potamogeton richardsonii; *retention; uptake; uptake sites; zinc.*

INTRODUCTION

Considerable research has been conducted relative to the uptake and translocation of nutrients and other elements by the roots and shoots of aquatic macrophytes (e.g., Toetz 1971, 1974, Denny 1972, DeMarte and Hartman 1974, Mayes et al. 1977, Best and Mantai 1978, and Ernst and Marquenie-van der Werff 1978). However, the measurement of uptake and loss rates of elements was not addressed in these studies. Cole and Toetz (1975) determined the half-saturation constants for NH_3 uptake by roots and rhizomes of *Potamogeton nodosus* and *Scirpus* spp. Considerable controversy exists about the relative role of the root and shoot systems as the site for element uptake in aquatic macrophytes. Sculthorpe (1967) advocates the roots as the main entry site, while Sutcliffe (1962) believes the shoots predominant. Denny (1972), after comparing growth rates of hydrophytes rooted in sand and mud, reported that root sorption appeared dominant, but that his experiments failed to demonstrate that roots were the exclusive site of entry.

Aquatic macrophytes are important constituents of many aquatic ecosystems which receive industrial effluents and thus must be considered in developing models of the cycling of effluent trace metal components within the system. The studies described below were designed to assess uptake and retention processes for Cu and Zn by two macrophytes, *Myriophyllum heterophyllum* Michx. and *Potamogeton richardsonii* (Ar. Benn.) Rydb.

Two possible scenarios for the exposure of macrophytes subjected to increased trace metal concentrations are (1) germination and growth of plants where high levels of trace metals are already present in the sediments and, perhaps, the water, and (2) germination and growth of plants in an uncontaminated pond which later receives effluents containing elevated levels of trace metals. In the experiments described below we have attempted to mimic the first scenario. Thus, the controversy about important routes of uptake has a direct bearing on our experiments since roots and shoots could both be sites of uptake. However, in the second scenario, the roots would probably play a lesser role until trace metals accumulated in the sediments.

METHODS AND MATERIALS

Field methods

Three ponds were selected to study Cu and Zn uptake and retention by macrophytes and sediments: (1) Gable Mountain Pond (GMP), (2) Quarry Pond (QP), and (3) artificial ponds containing QP sediments and Columbia River water. GMP is a 29-ha lake on the Hanford Reservation in south central Washington which receives low-level radioactive effluents from chemical operations and also contains high levels of trace metals in its sediments (C. E. Cushing, *personal observation*). Detailed limnological characterizations of GMP are in Cushing and Watson (1974) and Emery and McShane (1980). QP is a 5-ha pond formed from underground seepage after closure of McNary Reservoir on the Columbia River. It is approximately 50 km downstream from the Hanford Reservation and contains lower levels of trace metals in its sediments. Artificial ponds were located at the Department of En-

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TABLE 1. Mean concentrations of Cu and Zn in water (mg/L), sediments (μg/g dry mass), and macrophytes (μg/g dry mass).

	Gable Mountain Pond		Quarry Pond		Columbia River	
	Cu	Zn	Cu	Zn	Cu	Zn
Water	0.004 (2)*	0.01 (2)	...†	...†	0.003 (42)‡	0.016 (40)‡
Sediments	80 (45)	860 (45)	16 (45)	59 (41)	100 (1)	200 (1)
Potamogeton richardsonii	26 (1)§	182 (1)§	21 (1)§	40 (1)§	... ‖	... ‖
Myriophyllum heterophyllum	22 (6)	332 (6)	7 (1)§	32 (1)§	... ‖	... ‖

* Numbers in parentheses are number of measurements.
† Not measured.
‡ From Silker (1964).
§ Composite of six individual plants.
‖ Plants not indigenous in Columbia River.

ergy's Pacific Northwest Laboratories on the Hanford Reservation. Artificial ponds were used since we could not transfer radioactive plants and sediments (see below) from GMP to QP, an off-site, nonradioactive environment. Table 1 presents data on Cu and Zn levels in water, sediments, and macrophytes from the three study sites.

The experiments were conducted with sediments and two species of macrophytes, *M. heterophyllum* and *P. richardsonii*. In the sediment studies, sediments from the upper 12.7 cm of QP were carefully placed into a 30.5 cm by 30.5 cm cylindrical glass container. The container with sediments was transported and placed into GMP to assess any changes in Cu and Zn concentrations as a function of time. Concurrently, similar sediment samples were collected from GMP and placed in the artificial ponds. Three 1 × 10 cm cores were removed from each container at appropriate time intervals (more frequently during the initial portions of the 100-d period), divided into upper and lower halves, and analyzed separately. In the first macrophyte transplant study, approximately 150 plants of each species were collected from QP and replanted in GMP to assess changes in Cu and Zn concentrations. For the second study, a similar number of both macrophyte species from GMP were transplanted to the artificial ponds which contained about 20 cm of sediments collected from the upper 12 cm of QP and were filled with Columbia River water. Six individual plants were analyzed for each species from each transplant experiment at each sampling interval. Few plants failed to grow after being transplanted.

Laboratory methods

Copper and Zn concentrations in water were obtained using an APDC (ammonium-1-pyrrolidine dithiocarbamate) coprecipitation technique followed by energy-dispersive X-ray fluorescence spectrometry (Elder et al. 1975).

Plant shoots were analyzed after careful rinsing with distilled water to remove sediments, and separation of shoots and roots. Shoot and sediment samples were dried at 105°C prior to grinding and pressing into pellets for subsequent analysis by X-ray fluorescence spectrometry (Nielson 1977).

Quantitative methods

Nonlinear least squares techniques and an iterative computer program based on techniques in Draper and Smith (1967: Chapter 10) were used to examine the fit of several mathematical models to the data from macrophyte uptake and retention experiments. Linear regression models were used to describe results from sediment experiments. Examination of error mean squares and von Neumann ratios (von Neumann 1941), obtained after fitting several nonlinear models, led us to select a modified exponential (Mitcherlich) equation to model data from retention experiments and an exponential model to describe uptake data. To avoid computing problems while fitting the exponential model (it intercepts zero at $t = 0$), trace element concentrations at zero time were subtracted from the subsequent data, and after fitting the model, initial subtracted values were added to all observed and calculated values. The two equations we selected are appropriate when the mode of trace element transfer occurs in a closed system (heavy lines in the upper panel of Fig. 1 in Cushing et al. [1975]) and where either the plants or sediments were spiked (contained added radioactive or stable elements). We have assumed that when aquatic macrophytes from one location are transplanted to another with different Zn and Cu concentrations, then any resultant change is a measure of overall net rate of uptake or retention (and thereby half-time, or the time for 50% of the original concentration to be lost or gained). We believe these can be used to estimate the time of maximum or minimum Zn or Cu concentrations in macrophytes in situations where effluent discharges contain these elements. These experiments were not designed to determine individual rate constants between compartments, but instead to estimate the parameters describing overall uptake and retention kinetics. Individual rate estimates (Fig. 1) for Cu and Zn movement between compartments remain for experimental determination.

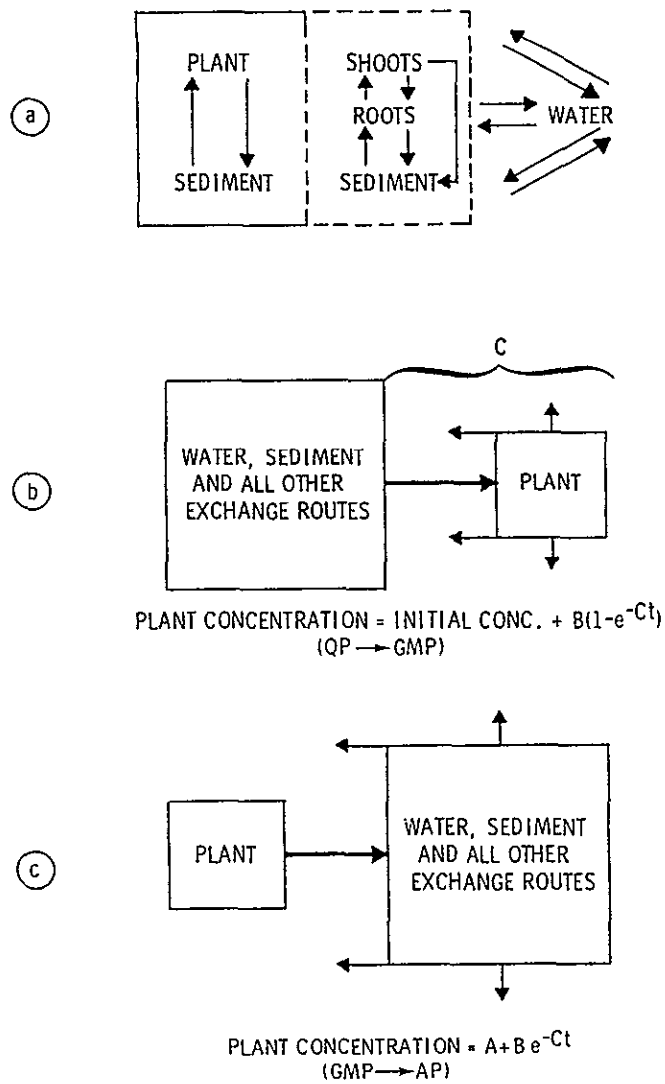

FIG. 1. Simplified model for trace metal transport among aquatic macrophytes, water, and sediments. (a) General scheme. (b) Plants grown in low Zn and Cu environments and transplanted to site with higher levels. (c) Plants grown in high Zn and Cu environments and transplanted to site with lower levels. The model parameters A, B, and C, can be interpreted in terms of rates (see Cushing et al. [1975]) for a closed, spiked system (see text). In these studies the uptake parameters B and C (Fig. 1b), can be thought of as the amount of trace metal accumulated (from $t = 0$ to infinity) and the rate of overall process, respectively. In Fig. 1c, the parameters represent the initial amount in plants (A and B), the final amount in plants (A), and the rate of loss (C).

TABLE 2. Calculated half-times (days) and a comparison of Zn and Cu concentrations (μg/g dry mass) predicted by the models in Figs. 3 and 4 with measured values in Table 1.

Plant source	Location during experiment	Element	Species	Concentration: Predicted (100-d)	Concentration: Measured value	Calculated half-time
QP*	GMP*	Zn	P†	191	180	1.3
			M†	304	330	5.0
		Cu	P	14‡	17‡	18.0‡
			M	23	21	4.7
GMP	AP*	Zn	P	90	40§	7.8
			M	244	32§	5.4
		Cu	P	10	21§	1.4
			M	17	7§	12.1

* QP = Quarry Pond: GMP = Gable Mountain Pond; AP = Artificial ponds, Columbia River water.
† P = *Potamogeton richardsonii*, M = *Myriophyllum heterophyllum*.
‡ Used model for loss from plant (Fig. 1c).
§ QP values: no data available for Columbia River plants.

RESULTS AND DISCUSSION

In the sediment transfer experiments, linear regression analysis revealed that only the slope of the line which described Zn concentration as a function of time, in the upper 5 cm of sediments transferred from QP to GMP, was significantly >0 ($P < .01$) over the 100-d study (Fig. 2). Calculated slopes were all positive (Cu about 0.02; Zn 0.07 to 0.45) which may suggest that Zn is the more mobile of the two elements under these conditions. Even though Zn concentration in the upper 5 cm of sediments from QP increased when transferred to GMP, GMP sediments did not lose Zn when transferred to Columbia River water in the artificial ponds. Instead, they increased slightly but the change in slope was not statistically significant, even though Zn concentrations were higher in GMP than CR water. It is possible that the chemical form of Zn was different in the two systems.

The results of nonlinear least squares fits of *P. richardsonii* and *M. heterophyllum* data to the mathematical models in Fig. 1 are shown in Figs. 3 and 4 respectively. Half-times obtained from these models (Fig. 1), as well as a comparison of the predicted concentration values with initially measured values, are in Table 2. Predicted concentrations calculated after 100 d of uptake for both plants and trace metals are in excellent agreement with measured values determined for resident plants (Tables 1 and 2). However, we could not compare predicted final concentrations of GMP plants transferred to the artificial ponds (retention) because these plants do not occur in the Columbia River. Predicted 100-d concentrations were not comparable to concentrations found in QP macrophytes (Table 2).

Respective half-times ranged from 1.3 to 18.0 d for uptake and from 1.4 to 12.1 d for retention experiments. The 18-d half-time obtained for Cu in the *P. richardsonii* uptake study is questionable because we fit the retention function, instead of the uptake model, to those data after removal of an apparently aberrant observation. Even though Cu concentrations in *P. richardsonii* grown in GMP and QP were similar (26 and 21 μg/g dry mass, Table 1), we found that only a retention function fit the data. Both the concentration data and the resulting model (retention instead of uptake) were surprising based on the "contaminated" status of GMP. Examination of all half-times shows

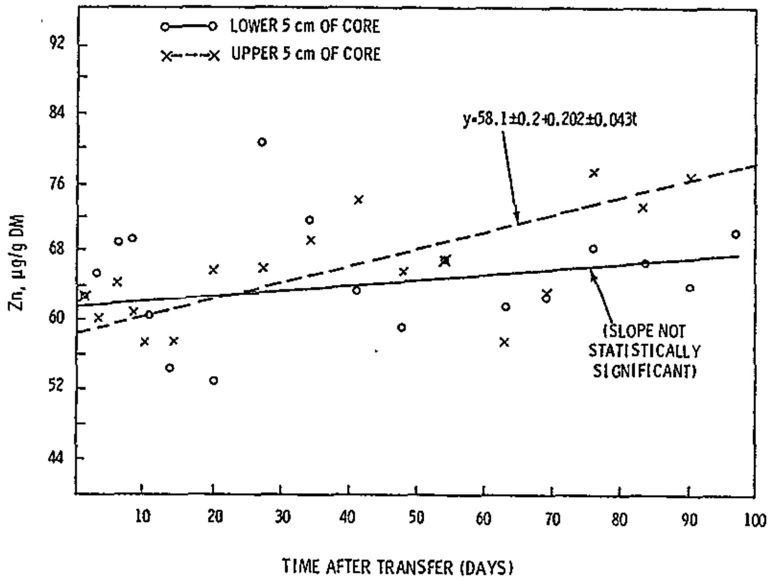

FIG. 2. Zn concentrations as a function of time for 1 × 10 cm cores of sediments transferred from Quarry Pond to Gable Mountain Pond.

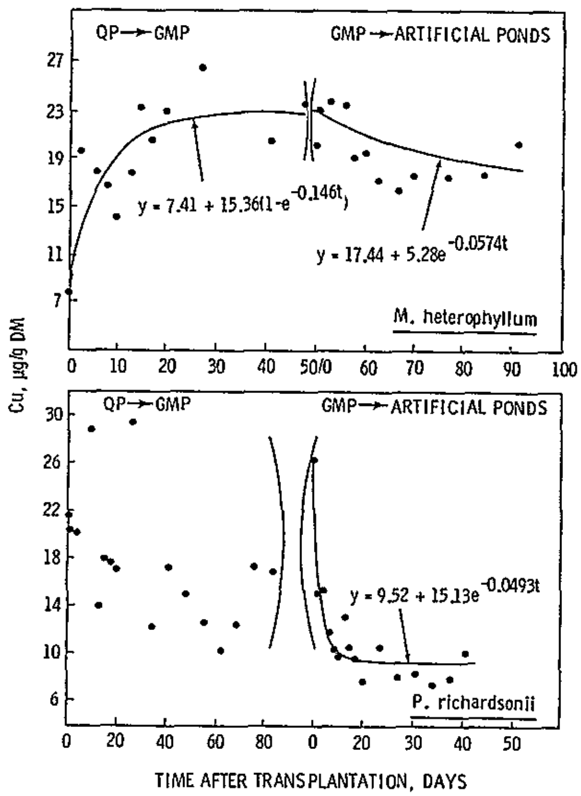

FIG. 3. Predicted and measured uptake and retention of Cu by *M. heterophyllum* and *P. richardsonii.*

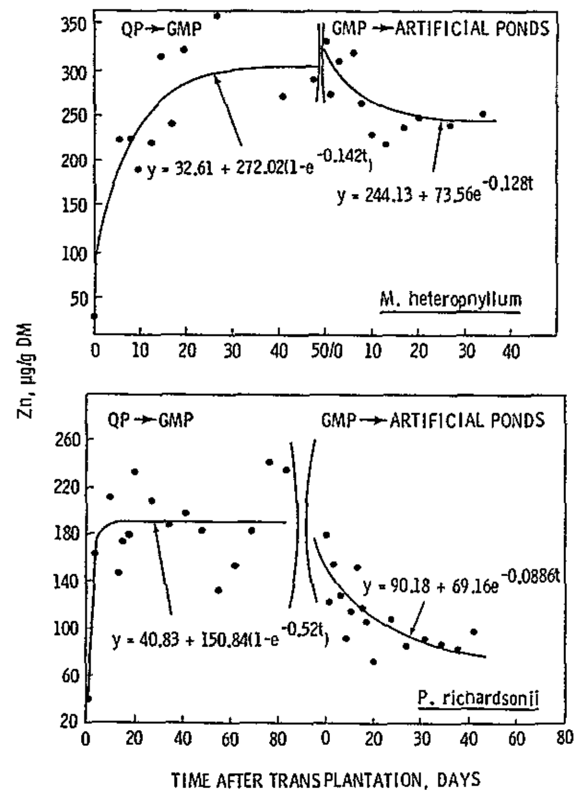

FIG. 4. Predicted and measured uptake and retention of Zn by *M. heterophyllum* and *P. richardsonii.*

little difference between plant species, uptake and retention, or trace metals. The largest numerical difference (12.1 and 1.4 d) for Cu in both macrophytes after transfer from GMP, probably is not statistically significant. However, the statistical methods used to test half-time differences depend on the validity of the linearizing approximation used to obtain error estimates (Draper and Smith 1967). Uptake and retention half-times averaged 3.7 d (with the aberrant value of 18.0 deleted) and 6.7 d, respectively. Many additional experiments would have to be conducted before such a small difference could be declared statistically significant.

Ratios of Cu and Zn concentrations determined in both plants obtained during uptake and retention experiments showed that *M. heterophyllum* contained about twice the Cu and Zn when compared to concentrations found in *P. richardsonii* (1.8 and 2.4 for Cu and Zn, and 1.4 for Cu during retention and uptake, respectively). Small and similar coefficients of variation (about 25%) for all three data sets showed that these observations were consistent. We speculate that Cu or Zn determined for one plant species may be useful in estimating the concentration in the other, even during the time of exposure to elevated and/or decreasing ambient levels in the sediments (at least under our experimental conditions).

Conclusions

These studies indicate that when increased concentrations of Cu and Zn are introduced into uncontaminated ponds, aquatic macrophytes (*P. richardsonii* and *M. heterophyllum*) can be expected to incorporate them into the vascular tissues. Initial movement from the water into the sediments is very slow, at least during the initial 100 d. Apparently, once the sediments accumulate the metals, uptake by these macrophytes is rapid (except for Cu in *P. richardsonii* transferred from QP to GMP). The converse is also true; macrophytes with high concentrations of Cu and Zn will rapidly lose Cu and Zn if increased environmental levels are somehow reduced. In nature, rapid changes in ambient water concentrations can be postulated, but it does not seem conceivable to change the sediment concentrations rapidly.

It should be remembered that the concentration changes observed in these experiments are the result of complex interactions among the plants, water, and sediments, including the possible change in chemical form of Cu and Zn. We have attempted to emphasize the root-sediment relationships in this paper; we have, however, also conducted similar transfer experiments in which the plants with root systems and sediments intact were transferred in jars to examine the shoot-water interactions. These studies indicated that the shoots were essentially inactive in uptake and loss of Cu, but were active in the exchange of Zn, although root uptake of Zn appeared predominant. Thus our results support the hypothesis of Sculthorpe (1967) that the roots are the main site of uptake, at least for Cu and Zn. Additional evidence from our experiments with root systems and sediments transplanted intact, however, supports the findings of Denny (1972), because roots were not the exclusive site of uptake for Zn.

Our Zn and Cu data do not exhibit the oscillations during uptake and subsequent loss as found for Pb and Cd in *Elodea* by Mayes et al. (1977). They were unable to estimate rate constants, probably due to the infrequent, tri-weekly measurement schedule. We have some limited data on Pb concentrations in *P. richardsonii* and *M. heterophyllum* which do not show rapid oscillations.

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